Short communication

EFFECTS OF LITHIUM ON Phaseolus vulgaris L.*

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ABSTRACT

The effects of lithium (Li) in growth media on *Phaseolus vulgaris* L. 'Bush Blue Lake 290' (snap bean) were studied under controlled environmental conditions. Concentrations of 4 to 12 ppm Li in the substrate produced toxic symptoms. The development of these symptoms is described and measurements of selected growth parameters presented. At 4 ppm Li, plant height, first trifoliate fresh weight and leaf area increased. At concentrations greater than 4 ppm stomatal diffusive resistance increased temporarily, indicating partial stomatal closure. It can be concluded that Li interferes with biomass accumulation and plant water relations.

INTRODUCTION

At high concentrations, lithium (Li) toxicity has been shown in plants. Whereas relatively lower Li levels have produced stimulatory effects. Although some information is available on the affect of Li on physiological processes, at present we know little about Li toxicity in plants.

The phytotoxicity of Li was first described by Gaunersdorfer [1]. Ravenna and Zamorani [2] demonstrated pronounced toxic effects to bean grown in solutions containing 250 ppm Li. More recently, Bingham et al. [3] reported slightly chlorotic leaves on dwarf red kidney bean in solutions maintained at 5 ppm Li. Plants grown in soil at 10 and 12 ppm Li exhibited leaf injury and 25% growth depression, respectively. Increases in plant height, dry weight, and seed yield were observed by Nakamura [4] in pea and barley at substrate concentrations of 2 and 19 ppm Li. Voelcker [5] found wheat shoot weight increased at 18 ppm Li in the substrate. Brenchley [6] showed a

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doubling of dry weight in barley grown in solutions at 0.02 ppm Li. Lithium-stimulated growth results have not been clearly described.

Lithium at 7 ppm in solution diminished the action of phytochrome on flower induction in *Lemna* spp. [7] and retarded the circadian rhythm of petal movement in *Kalanchoe blossfeldiana* [8]. Solutions containing 7 ppm Li also blocked thigmomorphogenesis in *Bidens pilosus* L. var. radiatus [9] and *Bryonia dioica* [10]. Louguet and Thellier [11] reported that 35 ppm Li significantly reduced the degree of opening of stomata and the speed of opening and closing in *Pelargonium* x hortorum.

Lithium is used in several industrial processes and, according to most recent statistics, apparent consumption increased 55% in the 7 year period, 1971-1977 [12]. This trend may lead to an increase in the release of Li into the environment. Critical dose-response designs for Li have not been conducted under controlled environmental conditions. Thus, we designed an experiment to follow the development of toxic symptoms of Li on *Phaseolus vulgaris* L. 'Bush Blue Lake 290' (snap bean). We also studied the threshold level for growth suppression and possible growth stimulation by Li. In addition, we monitored stomatal diffusive resistance (R_s) during the experiment to assess possible effects of Li on stomatal activity.

MATERIALS AND METHODS

P. vulgaris seeds were germinated in 240-ml Styrofoam [13] cups containing a growth medium of 1/3 peat-lite and 2/3 gravel (No. 16 crushed stone). Seedlings were transplanted at 7 days into 11-cm plastic pots containing the same mix. Sixteen plants were randomly arranged in an environmental growth chamber of the Southeastern Plant Environmental Laboratories at North Carolina State University, Raleigh, NC [14]. The growth chamber was maintained at $26/22^{\circ}$ C (day/night) temperatures at approximately 77% relative humidity. A combination of fluorescent and incandescent lamps provided 9 h of light daily at a quantum flux density of $600\,\mu\rm Einstein\,m^{-2}\,sec^{-1}$ (400–700 nm). Pots were watered daily with nutrient solution [14].

Twelve days after transplanting, as the first trifoliate was almost fully expanded and the third was just developing, the substrate was saturated with one of four lithium nitrate (LiNO $_3$) solutions. These solutions gave initial concentrations of 0, 4, 8, and 12 ppm Li in the substrate. After the application, plants were returned to the standard watering schedule. They were observed each day for signs of visible injury. Also, $R_{\rm s}$ was measured on both leaf surfaces 1 day before and for 6 days after treatment with a diffusive porometer (Lambda model LI-60, horizontal sensor). The porometer was centered on one-half of the central leaflet of the first trifoliate for the upper surface and on the other half of the central leaflet for the lower surface.

Plants were harvested 8 days after treatments. Plant heights, leaf areas,

and fresh and dry weights of leaves, stems and roots were measured. Plant material was oven-dried for 3 days at 60°C for dry weight determinations. The plastochron index (PI), a measure of the physiological age (20 mm reference length), was determined at harvest [15]. Data were subjected to multiple regression analysis, using the Statistical Analysis System [6].

RESULTS

Toxic symptoms were most pronounced on the first trifoliates and were quite evident for the 8 and 12 ppm Li treatments. Control plants appeared healthy and robust. Marginal chlorosis was evident on the first trifoliates at 8 and 12 ppm Li 1 day after treatments. This was accompanied by a marked flaccidity of and grey-green lesions on the second trifoliates. Vascular bundles of stems and leaf petioles and major veins of the first trifoliates appeared red (possibly due to anthocyanin accumulation) 2 days after the 12 ppm treatment was initiated. After 3 days, the 12 ppm Li treated plants exhibited abnormal, nastic, development of the laminae of the third and fourth trifoliates. Plants treated with 4 ppm Li were larger than controls even though their first and second trifoliates exhibited very slight toxic symptoms. Visible effects of Li on plants 1 week after treatment are described in Table 1.

Primary leaf areas and fresh weights were not affected by Li. The 4 ppm Li treatment significantly increased the leaf areas of the first (Fig. 1) and second trifoliates. First trifoliate fresh weight responded similarly with the greatest fresh weight at 4 ppm as shown in Fig. 1. Second trifoliate fresh weight decreased at 8 and 12 ppm Li with no increase at 4 ppm Li. Only

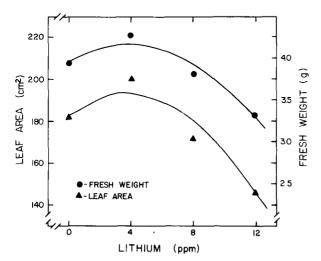


Fig. 1. Effects of lithium on *P. vulgaris* first trifoliate leaf area (\triangleq); $y = 183.34 + 5.27x - 0.716x^2$; and fresh weight (\bullet); $y = 3.98 + 0.099 - 0.013x^2$. Each point represents the mean of four observations.

TABLE 1
LITHIUM TOXICITY SYMPTOMS IN P. Vulgaris ONE WEEK AFTER TREATMENT

Leaf	Li concentration (ppm)		
	4	8	12
Primary	_	Marginal chlorosis with tip and partial marginal necrosis	Overall chlorosis and interveinal necrosis
Trifoliate:			
first	Trace tip and partial marginal chlorosis	Interveinal chlorosis and tip necrosis	Overall chlorosis and interveinal necrosis
second	Trace overall chlorosis	Interveinal chlorosis and partial tip necrosis	Interveinal chlorosis and tip necrosis to overall chlorosis, interveinal necrosis, and abnormal leaf development
third	_	Trace tip and marginal chlorosis	Tip chlorosis to tip necrosis with abnormal leaf development
fourth	_	-	Trace marginal chlorosis, some with tip necrosis
fifth	_	_	_

12 ppm Li decreased leaf area and fresh weight of the third trifoliates. There were no significant Li effects on fourth or fifth trifoliate leaf areas or fresh weights.

Root fresh weight decreased as Li concentrations increased. Similarly, stem fresh weight decreased as Li concentration increased. Analysis of shoot height data produced a quadratic relationship with the tallest plants at 4 ppm Li and the shortest at 12 ppm Li. Dry weights of roots, stems and leaves were negatively correlated with Li treatments (Fig. 2). The physiological age of the plants was not significantly affected and had mean plastochron indices of 6.91, 6.88, 6.77, and 6.98 for the 0, 4, 8, and 12 ppm Li treatments, respectively.

Measurements of R_s for both adaxial and abaxial surfaces were computed for the total leaf. Total leaf R_s was not affected at day 1 after treatment at any Li level (Fig. 3). However, R_s increased at 8 and 12 ppm Li on days 2 and 3 and at 12 ppm Li on day 4 after treatment. We observed no significant differences among treatments at 5 days. Results were similar when the upper and lower surfaces were analyzed individually.

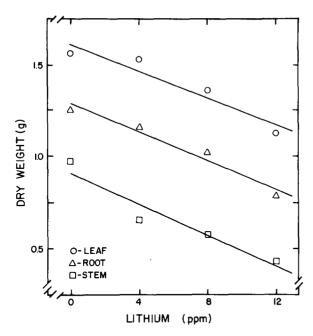


Fig. 2. Effect of lithium on *P. vulgaris* leaf (\bigcirc); y = 1.62 - 0.04x; root (\triangle); y = 1.29 - 0.04x; and stem (\square); 0.91 - 0.04x; dry weights. Each point represents the mean of four observations.

DISCUSSION

Lithium produced chlorosis of leaflet margins extended interveinally towards the base of the leaflet with time. Necrosis occurred at higher Li concentrations, mainly at leaf tips and margins, and also proceeded towards interveinal portions of the leaflet. Production of leaf lesions and reddening of vascular tissue may be explained by high local Li concentrations. The accumulation of anthocyanins is a well known symptom of stress in plants. Haas [17] explained the abnormal expansion of newly formed leaflets in experiments with *Citrus* spp. Differential deposition of Li between marginal and interveinal regions may allow cell expansion along veins while necrosis occurs marginally.

The first and second trifoliates were affected most by Li and new growth was progressively less affected. Kent [18, 19], using wheat, suggested that Li accumulated more in expanding or recently expanded leaves, became immobile, and was not retranslocated. Leaves with growth stimulation at 4 ppm Li also exhibited very slight chlorosis. Kent [18] predicted that growth stimulation could be produced in one region of a plant while a toxic effect could occur in another due to local differences in Li concentrations.

The increase in R_s suggested that Li does, in fact, affect the stomatal apparatus, as was previously reported [11], and either prevents or reduces

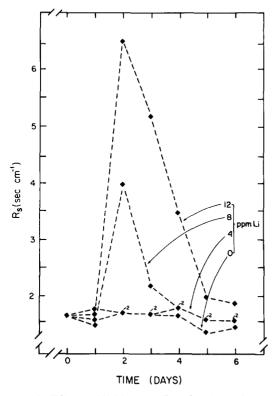


Fig. 3. Effect of lithium on P. vulgaris total stomatal diffusive resistance $(R_{\rm s})$ for the 0, 4, 8, and 12 ppm Li treatments with time. Each point represents the mean of four observations.

the opening of stomata. Besides reduced transpiration rates, net photosynthesis would also likely be reduced. However, this effect is temporary and plants resume normal stomatal regulation after a few days. The degree as well as the rate of recovery depends on the Li concentration. Several investigators have proposed that Li perturbs several regulatory processes because it interferes with other alkali metal ions and plasmalemma-mediated ion fluxes [7—10], including the stomatal mechanism [11].

Our study clearly demonstrates that Li can be toxic or stimulatory to the growth of *P. vulgaris*, depending upon its concentration. Reductions in biomass are accompanied by characteristic visible symptoms. Certain toxic symptoms can appear even when growth is stimulated. Lithium increased stomatal diffusive resistance but, within the range of Li and time levels studied, this increase was temporary and the rate of recovery was dependent upon Li concentration.

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